

end of the block is wet in solution no. 1, a piece of paper with the desired data is stuck on and washed over with solution no. 3.

In conclusion I may say that I have tried nearly all the methods employed in celloidin manipulation and have succeeded best with the above, which is largely a combination or modification of methods already known.

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The Collodion method in botany.

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In any original investigation in the field of histological botany the worker is at the outset confronted with the all important question of how to prepare the material so that it may be available in the best possible condition for thorough systematic observation and study. The old method of making freehand sections in careful investigations is now gone by and the best histologists look for some substance with which to infiltrate the tissue and bring it into a condition where uniform serial sections can be made. Many substances have from time to time been offered and met with varying success. Some of the more important ones that have been tried are gelatin, gelatin soap, gum, paper, shellac, wax, gum arabic, soap, paraffine and collodion. The last two substances have seemingly supplanted the others and indeed they seem to offer all the advantages that can be secured by any of the others.

The paraffine method as applied to plant tissue was published by Dr. Moll in the *BOTANICAL GAZETTE* of January, 1888, and later many modifications of it have been given and the method much improved. The method has been quite extensively used, but is very long and quite disagreeable to manipulate, often requiring 10 or 12 days to bring the tissue into proper condition for sectioning. For proper infiltrating with paraffine a temperature of from 45° to 50° C. is required, and for some of the more delicate tissues this in my hands has proved fatal. The method is admirably adapted, however, for many tissues that can be held in position for sectioning in no other way, as is the case with mature seeds or woody stems. The collodion method is now coming into general use for nearly all kinds of plant tissue. For the use

of collodion for infiltrating we are indebted to Duval who first published his results in the *Four. de l' Anat.*, 1879, p. 185. A little later Merkel and Schiefferdecker suggested the use of celloidin which is merely a patent collodion. This appeared in the *Archiv. f. Anat. u. Phys.*, 1882, p. 200. Some discussion then arose regarding the relative merits of each, but it is generally agreed that one has little or no advantage over the other. The method as applied to plant tissue is a comparatively new one. In 1884 we find a short description of it in Strasburger's Botany and some few modifications of it have since been offered.

In the histological laboratories at Cornell University, under the direction of Professors S. H. Gage and W. R. Dudley, I have tried the method with its various modifications on many different kinds of plant tissue, and find the following operations to be in every particular the best to obtain uniformly good results:

The tissue to be treated is first dehydrated and hardened in alcohol. For this purpose a Schultze's apparatus is of the first importance; in fact I have found some tissues that could be hardened in no other way without shrinking. The results fully warrant the statement that no one engaged in histological botany can afford to be without such an apparatus. Many forms of it have been suggested, but for most plant tissues one which will be found very convenient can be made by fitting into a Whitall-Tatum museum jar a rack in which several dehydrating tubes can be supported at any desired level in the alcohol contained in the jar. In place of the parchment diaphragm usually used I recommend the use of chamois skin. With such an apparatus, from 12 to 25 hours is sufficient for hardening and dehydrating any plant tissue. In using the apparatus the tissue should be packed closely in the dehydrating tube, and enough 50 % alcohol added to just cover it. The tube is then sunk in the 95 % alcohol in the jar until the two liquids are at a level. The strength of the 95 % alcohol can be kept up by adding to it, from time to time, some calcium chloride. This will not in any way injure the alcohol. The tissue is taken from the 95 % alcohol and placed in a 2 % solution of collodion, made by dissolving 2 grams of gun cotton in 100 cc. of equal parts of ether and alcohol. In this it is allowed to remain from 12 to 24 hours, depending on the nature of the tissue, 24 hours being enough for the very firmest. It is then transferred to a 5 % solution, or the 2 % solution is allowed to evaporate until it is of the consistency of the 5 % solution. The former method

is more uniformly satisfactory. The tissue is allowed to remain in this solution about 12 hours. It is then taken out and arranged in position on a cork or block of wood of convenient size to fit in the jaws of the microtome. It is not necessary that the corks be previously soaked in collodion. By means of a camel's hair brush the material on the cork is covered with successive layers of thick collodion until it is quite enclosed in the mass, allowing each coat to dry slightly before applying the next. After the tissue is covered it is allowed to harden in the air a few minutes and then placed in about 80 % alcohol to harden. Much difference of opinion exists regarding the proper strength of alcohol to use for hardening the collodion, but 80 % answers very well, and the tissue can be kept in it a long time without deteriorating. After a few hours the collodion will be hard enough to section. For sectioning any sliding microtome will answer the purpose, but one especially adapted for this work will enable one to incline the object (which can be clearly seen through the collodion) in any desired position and to take sections in any desired plane. It is also necessary that the sections be removed with long sweeping cuts, since direct cross cutting would tear them. The sections should be kept covered with alcohol while being removed, and then floated from the knife to the slide. The slower the section is cut the better it will usually be. Serial sections can be arranged in their proper place on the slide. For fixing the sections some dry ether ether vapor is blown upon them. A very convenient form of apparatus for the purpose can be made by fitting two tubes into a wide-mouthed bottle as in making a wash-bottle, except that the entrance-tube dips below the surface of the ether and the exit-tube is above the ether. The inner end of the exit-tube should be expanded into a bulb, in which calcium chloride is placed to dry the vapor as it passes out. The ether vapor dissolves the collodion and fastens the sections to the slide. The sections are then washed with water, stained, the surplus stain washed off with water, the sections dehydrated with alcohol, cleared and mounted in balsam. For clearing, a mixture used by Prof. Gage for animal histology will be found to work admirably. It consists of 3 parts of turpentine and 2 of carbolic acid. It clears quickly and will not injure the most delicate tissue. For staining, one must use that which seems best adapted for his purpose, but for general study hæmatoxylin seems especially adapted to collodion sections.

Some difficulty may arise in cutting sections which have

in them free parts. It sometimes happens that they become detached from the collodion and float away. In this case the section can be collodionized as was first suggested by Dr. Marks. This is done by coating the tissue before each section is cut with a thin coat of 1 % collodion, using a camel's hair brush for the purpose. Then draw the knife across the tissue very slowly, keeping alcohol dripping on it while the section is being cut. In this way beautiful longisections of large compound pistils can be obtained in which the sections of ovules, though not held in place by the placenta, will, nevertheless, remain in their proper position and perfect serial sections of each ovule obtained.

Care should be taken that none of the sections be cut before collodionization, for although it may not always be necessary to keep the parts in place, yet it is always a safeguard against their displacement. The method as given is found to work admirably on very delicate meristematic tissue. No heat being required the most delicate of tissue will not shrink. Then, too, the shortness of the method commends it to general use. I find that 2 days or even 36 hours is sufficient and is even better than a longer time to go through the whole operation of hardening, infiltrating and sectioning nearly all kinds of plant tissue. The material may apparently be left in the thick collodion indefinitely without deteriorating. The sections after being cut can be handled with a camel's hair brush without danger of breaking. By a little experience one will find that the method as given will enable him to bring to his hand material with which to pursue with certainty any investigation in histological botany.

Cornell University.

A biographical sketch of J. B. Ellis.

F. W. ANDERSON.

(WITH PORTRAIT.¹)

The subject of the present sketch was born at Potsdam, N. Y., January 21, 1829. He evinced a remarkable fondness for study at an early age, and the time not spent at school or at work on his father's farm was devoted to reading. At the age of sixteen he taught the winter school at Stockholm, St. Lawrence county. Here the lad received for his services ten dollars a month and "boarded around." Five of the ten dollars was paid in cash, the other five was

¹By mistake of binder the portrait of Mr. Ellis was bound with the October GAZETTE.